Republic of Iraq
Ministry of Higher Education
& Scientific Research
University of Al-Qadisya
College of Veterinary Medicine

Toxopathological effect of arsenic trichloride in white rats

A Graduation Project Submitted to the Department Council of the Internal and Preventive Medicine-College of Veterinary Medicine/ University of Al-Qadisiyah in a partial fulfillment of the requirements for the Degree of Bachelor of Science in Veterinary Medicine and Surgery.

By
Ali Nadhem Sharif

Supervised by

Atiaf Ghanim Rhyaf

2018 A.D. 1439 A.H.

فَتَعَٰلَى ٱللَّهُ ٱلْمَلِكُ ٱلْحَقُّ ۗ وَلَا تَعْجَلْ بِٱلْقُرْءَانِ مِن قَبْلِ أَن يُقْضَىٰٓ
إِلَيْكَ وَحْيُهُۥ ۖ وَقُل رَّبِّ زِدْنِى عِلْمًا ﴿١١٤﴾

من سورة طه

Certificate of Supervisor

I certify that the project entitled **(Toxopathological effect of arsenic trichloride in white rats)**

was prepared by Ali Nadhem Sharif
under my supervision at the College of Veterinary Medicine / University of Al-Qadissiya.

Supervisor

Dr.Atiaf Ghanim Rhyaf
Dept. of pathology and poultry disease
Coll. Of Vet.Med./ Univ. of Al-Qadissiya.

22 / 3 / 2018

Certificate of Department

We certify that **Ali Nadhem Sharif**
has finished his/her Graduation Project entitled **(Toxopathological effect of arsenic trichloride in white rats)**
and candidate it for debating.

Instructor

Dr. Muthanna H. Hussain

22 / 3 / 2018

Head of Dept of Int. and Prev. Med.

Dr. Muthanna H. Hussain

22 / 3 / 2018

Summary

Arsenic is an element spreading in the environmental, water and food could contain contaminantby Arsenic,and it is considerednational environmental health problem.Arsenic use in making the pesticides. Also, the arsenic present mixed with the coal in mining.The nephrotoxicity,The cardiovascular abnormalities, neurotoxicity,and diabetes mellitus, will occur when the body exposed to arsenic exposure. Furthermore,arsenic exposure couldaffectthe liver tissue and its function and causeshepatotoxicity. The present study shows mechanisms and pathogenesis of arsenictoxicity**.**

List of Contents

List of Figures (Optional)

figure	Organ	diagnosis
1	Liver	Histopathological section of liver shows thenormalarchitecture of theliver.x10 H&E(46)
2	Liver	Histopathological section of liver treated with arsenic show necrosis and inflammatory cellsX10H&E(46)
3	Kidney	:Histopathological section of kidney shows thenormalarchitecture of kidney .X10 H&E(46)

4	**Kidney**	**Histopathological section of kidney treated with arsenic show necrosis and inflammatory cellsX10 H&E(46)**
5	**Brain**	**Histopathological section of brain of rat treated with arsenic show degeneration and necrosis of neoron.X40 H&E(47).**
6	**Brain**	**Histopathological section of brain of rat treated with arsenic show vaculation of microglia .X40H&E.(47).**

CHAPTER ONE

INTRODUCTION

1-Introduction:

Arsenic is element dispensed in the environment in land, water and air (1).Arsenicenters the food chain by fertilizers, pesticides, rivers, rain, groundwater, and wastes discharge (2).Chronic Arsenicexposure has arelationship with cancer of the lungs, liver, and bladder, also causes impair in the renal and nervous tissues (3).It causes a great problem in the environment through by wide distribution in nature. It causes anexcess in the produce of H2O2 which causes oxidative stress (4),and cause damage to all types of molecules such as proteins, lipids, and nucleic acids(5)Arsenic affects the mitochondrial enzymes, inhipition the cellular respiration, and causes cellular toxicity. It can also substitute phosphate intermediates, which could theoretically slow down the rate of metabolism and interrupt the production of energy. (6)(7) Observed a positive correlation between arsenic concentration and lipid peroxidation levels in liver, kidney, and heart. Overproduction or an ineffective elimination of ROS may induce oxidative stress.Many Studies document that arsenic generates ROS are constantly produced in the brain in vivo(8),and it is induced pathological changes through oxidative DNA damage in the brain tissues in vivo and that cerebral and cerebellar cortex neurons seem to be the major targets of arsenic neurotoxicity.

The present study aimed to assess the effects of inorganic arsenic onsome organ in white rats.

CHAPTER TWO

REVIEW OF LITERATURES

2-Literature Review:

The heavy metals Exposure is a common problem overworld; this problem occurs in the drinking of contaminated air, water,and food. The arsenic is organic, inorganic element distributed innature(9).

1-2- History:

Arsenic used as a homicidal agentalso is used in some consumer products chemotherapeutic agent and pesticide.Late 18th century,the arsenic toxicity documented in Fowler's solutions, which was discovered in 1786.The potassium arsenite (1%) was used in the treatment of many diseases, such as malaria, asthma,syphilis, eczema, psoriasis,andchorea(11). Also, the Arsenic used in thetreatment of cancer, skin disease and breast cancer (12). In the 1950s arsenic use in organic pesticides where it began and continued into nowadays.

2-2-Physical properties:

Arsenic trichlorideor sometimes is called arsenous chloride. Arsenic trichloride iscolourless oils, sometimes take yellowcolour(13). It is causedirritating to the eyes, mucous membrane andskin. Arsenic trichloride is very toxicif it takes by ingestion and inhalation (14).

2-3- Chemical properties:

If Arsenic(III) oxide mixed with hydrochloric acid will result in Arsenic trichloride, it is preparation methods then submitted to the distillation. It appearslike many of forms:

1- Yellow (molecular non-metallic).
2- Several black and grey forms (metalloids) are a few that are seen(15).

2-4- toxicity:

2-4-1- Mechanism of toxicity:

Arsenic exists in the environment as pentavalent (As^{5+}, arsenate) and trivalent (As^{3+}, arsenite) forms(16).The arsenic is stored in many of organs such as kidney, heart,and kidney. Also, some studies documented arsenicin little amount nervous and muscletissue(17). The arsenic accumulated in tissues and associated with some diseases such as neurotoxicity, cardiac dysfunction, diabetes,and cancer. Arsenic can inhibitmore than 200 enzymes like DNA synthesis and cellular energy enzymes etc. (18). Arsenic compounds targeted enzymesin the mitochondria and causes impaired tissue respiration. resulting in the changes in enzymes pathway (19). Also, this supported by the fact that arsenic intoxication alters the activities of enzymes involved in cellular glucose uptake, gluconeogenesis, fatty acid oxidation and production of glutathione (20).

2-4-2-Physiological effect:

There are physiological effects of arsenic are complicated. Arsenic toxicitycauses negative health effects, cancer and some other diseases in people(21).The poisoning by acute arsenic is causedabdominal pain, vomiting,andnausea(22). While arsenite contaminated water and accumulate in the tissues through drinking of water and leading to several diseases such as diabetes, ischemic heart diseases, hypertension, atherosclerosis,nephrotoxicity, hepatotoxicity, and cancer of the lung, bladder and skin (23).

2-4-3- Pathological effect:

Humans and animals could expose to contaminated soil, food, water, air by arsenic (24,25). Cancer could occur and general severe diseasesassociated with CNS, kidney, liver and diabetes diseases (26)(27,28). Recently, some of theepidemiological studies in animal and human indicated an association between arsenic exposure and diseases [29-30]. [31,32] have indicated some tissues changedsuch ashepatic and heart by sodium arsenite. SodiumarseniteExposure showed degeneration and separation of muscle bundles and myocardiumhaemorrhages. The tissues of the lung after arsenic exposure showed anabnormal alveoli spaces and with abnormal alveoli cell [33]. Also, high concentrations Deposition of arsenic in thetissue of the hair, nails, kidney, and liver[28]. Furthermore, some epidemiological studies showed anassociation between kidney and liver diseases and arsenic exposure [28].

2-4-3-1- Pathological effect on liver:

Because of its unique metabolic functions and related to the gastrointestinal tract, theliver is an important target of toxicity to xenobiotics. Many types of research demonstrate That histopathological examination of liver sections after 24 hr of arsenic exposure show ballooning degeneration, necrosis in hepatic parenchyma with mild to moderate fatty change and lymphocytic infiltrate in the liver. (34) Also observed mild to moderate sinusoidal dilation characterized by widening of hepatic capillaries which may include the whole lobule or mostly in the central, periportal, or medial area, (35) showed moderate degeneration and. Besides that, there is infiltration of inflammatory cells(Fig2). The necrotic changes occurred as arsenic cause hepatocellular damage by producing increased reactive oxygen species which eventually disrupts the membrane system (36)

2-4-3-2- Pathological effect on kidneys:

The kidneys are the target organ due to its involvement in *vivo* biotransformation and elimination. Very few clinical cases of toxicity are reported in humans. Most clinical cases of toxicosis are reported in animals, especially cattle and dogs, by accidental exposure, but all species of domestic animals can be affected **(37).**

Histological alterations in kidney were observed in rats exposed to arsenic; the kidney showed changes in proximal tubular cells characterizedby swollen of tubular cells (38)(Fig 4). Also (39) estimated the oxidative DNA damage and pathologic changes in kidney tissue of mice treated with As2O3. Histopathological lesions recorded as cell swelling, tubular dilatation, lymphocytic infiltrations, loss of cell to cell contacts and loss of brush border in the epitheliums of proximal convoluted tubules.

2-4-3-3- Pathological effect on Brain:

Arsenic is a toxic heavy element and causes severe neurotoxic effects. Arsenic can accumulate in the brain and causes nervous diseases. Also, the arsenic is associated with thegeneration of the reactive oxygen species. Wherever, the mitochondria areimportant to thetarget of arsenic toxicity. (40)Reportedarsenicaccumulates in thebrain (41). Moreover, exposure tocadmium and arsenic causes impairs thedevelopment of the myelin and axon in rat brain [42]. The arsenic exposuredevelops neurotoxicity and causes disorder in behaviour[43].Also,it causes alterations in dopaminergic in rats [44]. Despite the line of reports on arsenic mediated brain disorders, none has evaluated the histological changes in thebrain by arsenic. In the current report, we have found edema, intracellular space, as well as anedematous change in arsenic-exposed

Chronic arsenic exposure increase of oxidative stress in therat'sbrain [45]. Therefore, we can assume that oxidative stress that caused by arsenic have agreat role in rat brain (Fig 5&6).

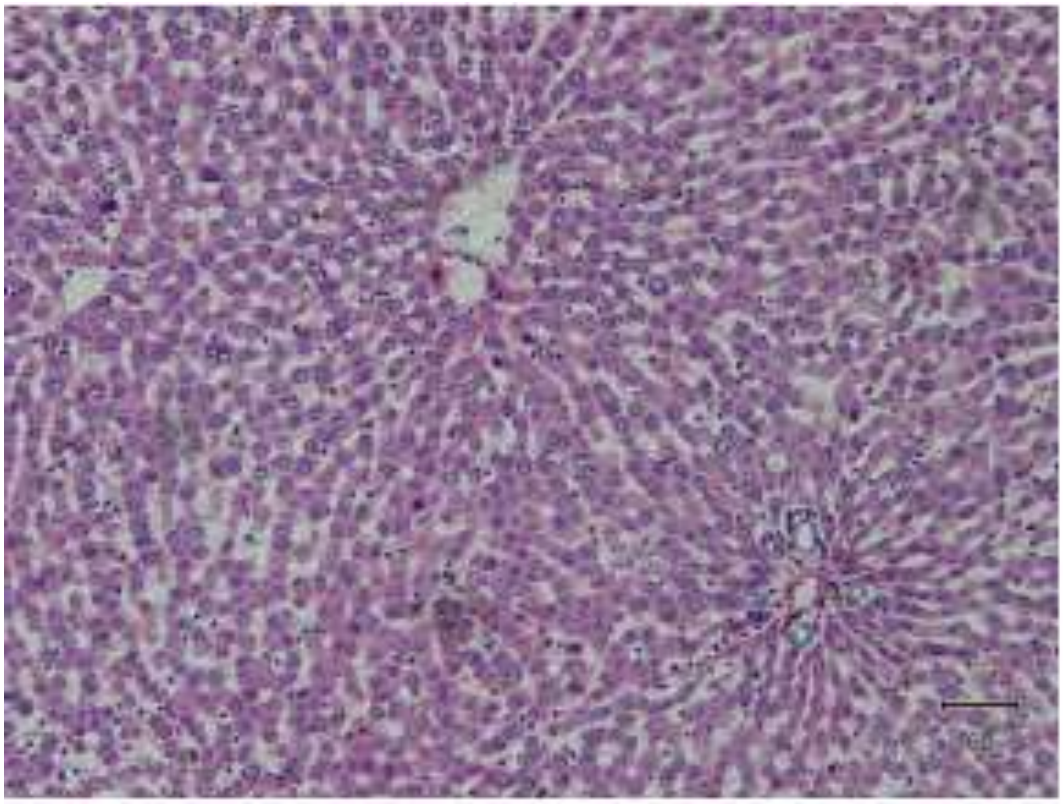

Figure1:Histopathological section of liver show normal artitucture of liver.x10 H&E(46)

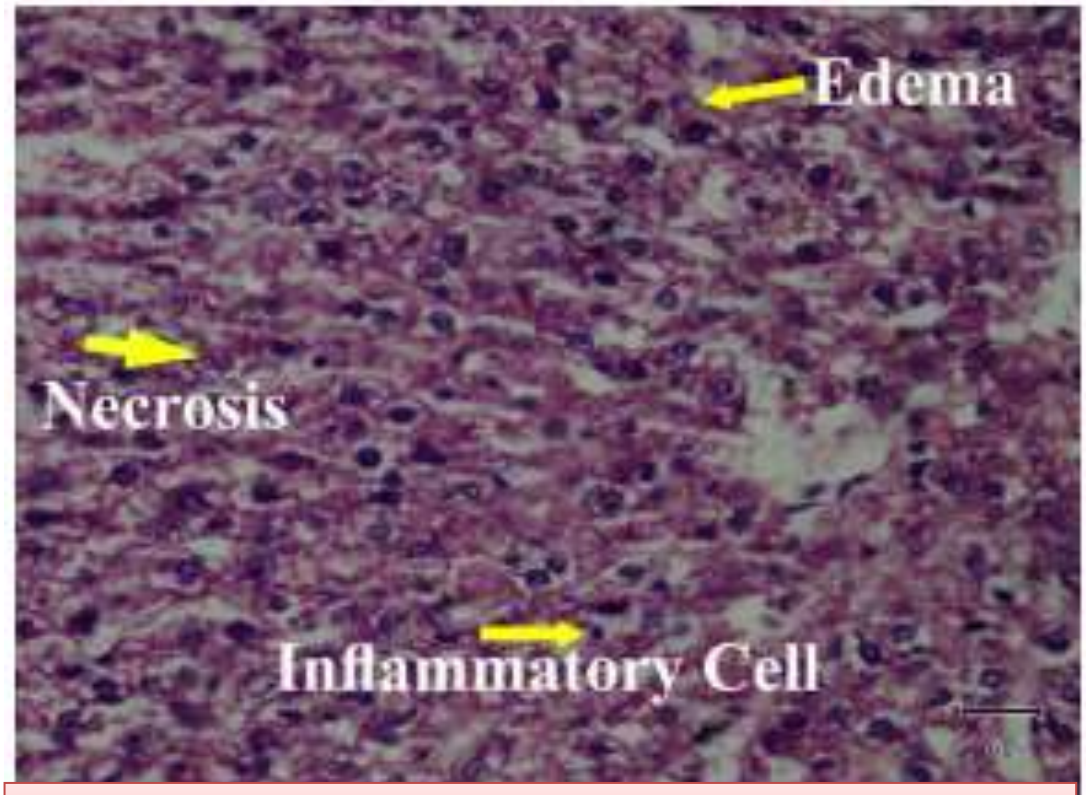

Figure2:Histopathological section of liver treated with arsenic show necrosis and inflammatory cellsX10H&E(46)

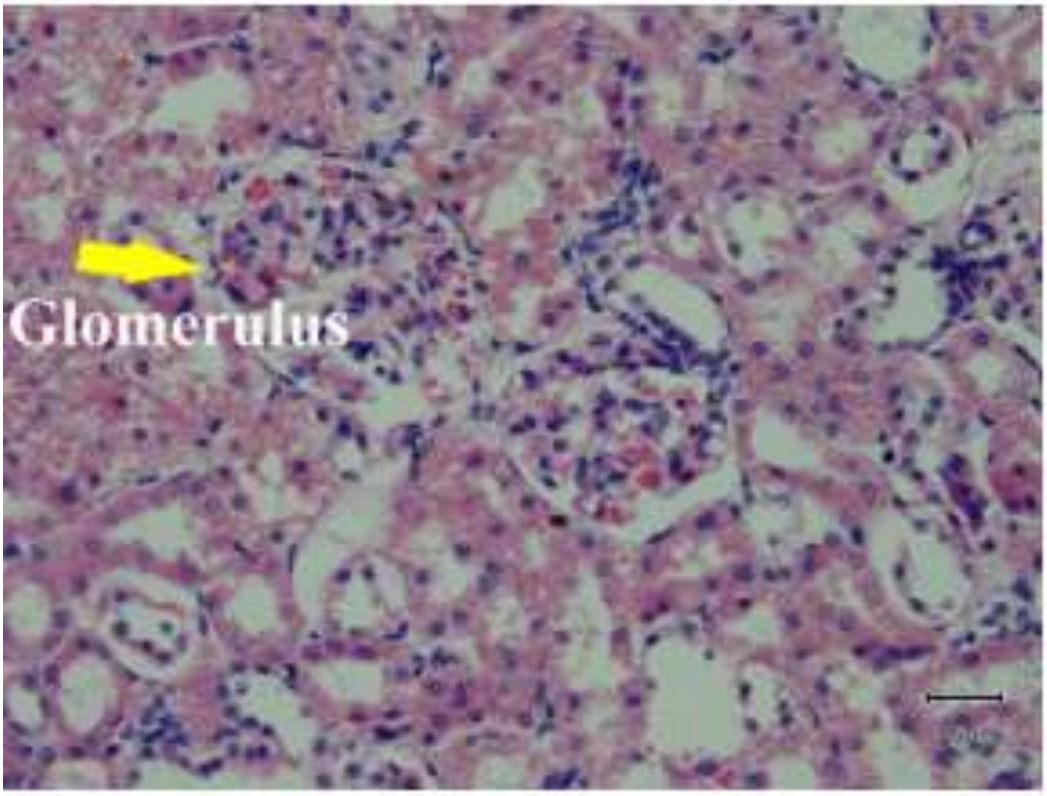

Figure3:Histopathological section of kidney show normal artitucture of kidney .X10 H&E(46)

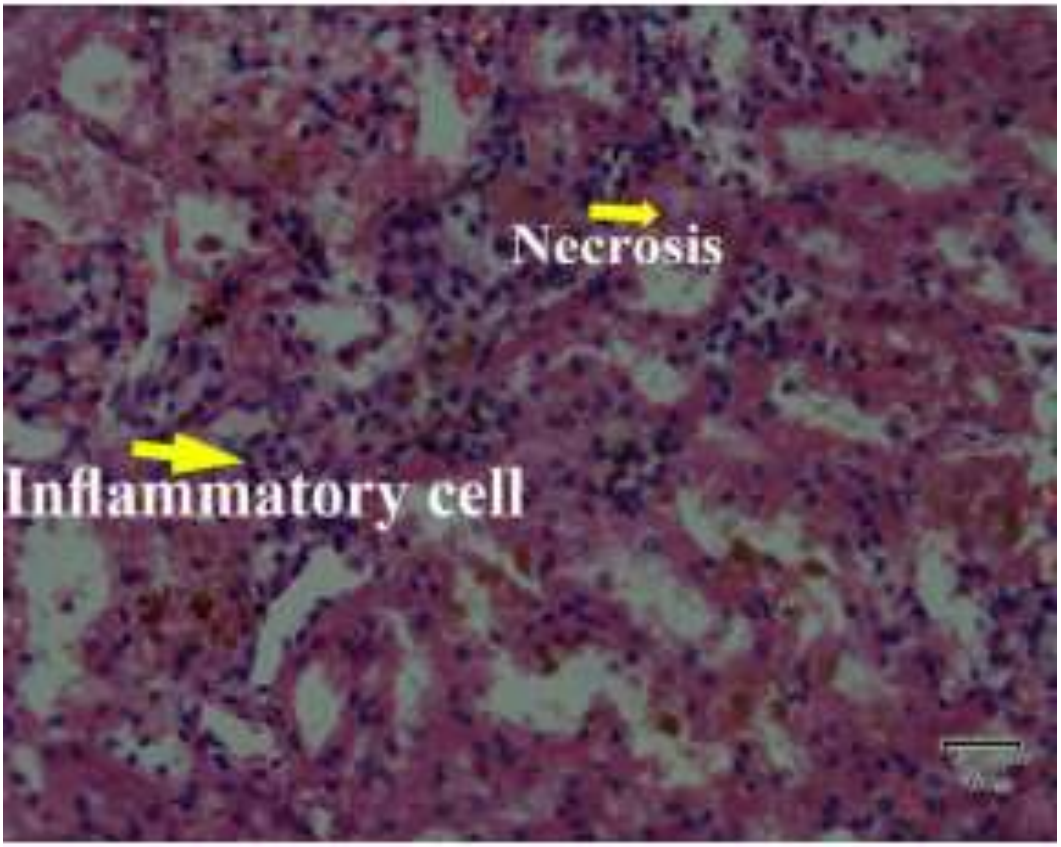

Figure4: Histopathological section of kidney treated with arsenic show necrosis and inflammatory cellsX10 H&E(46)

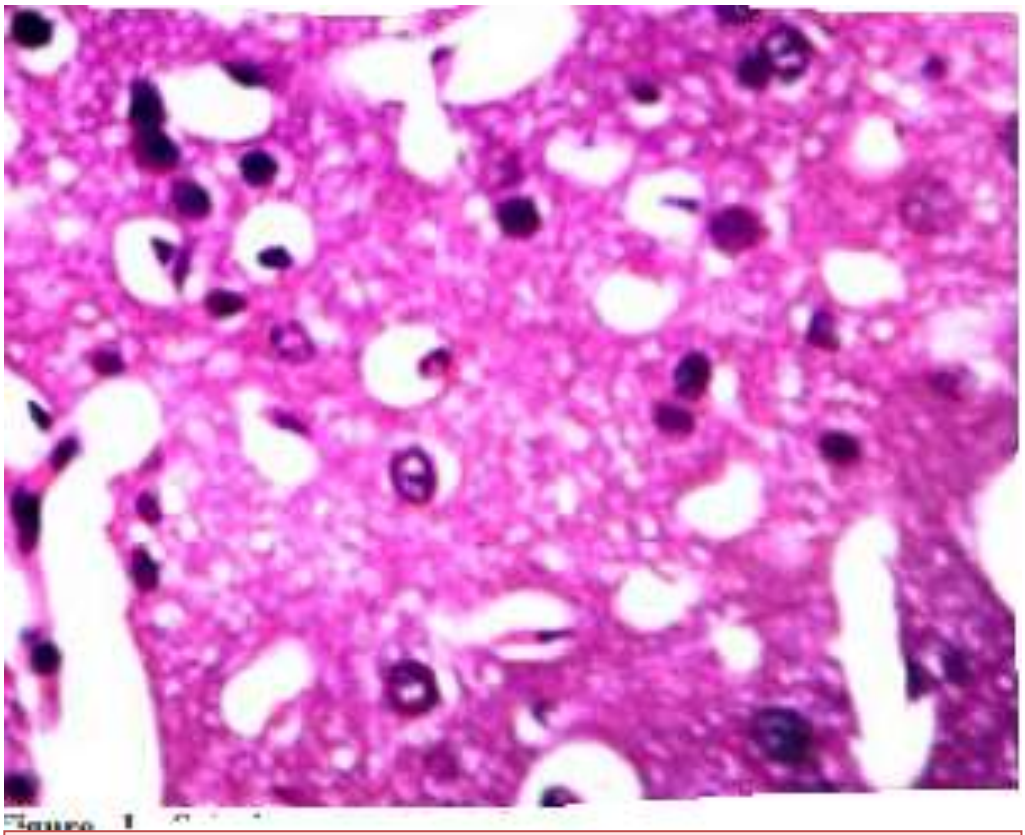

Figure6: Histopathological section of brain of rat treated with arsenic show vaculation of microglia .X40H&E.(47).

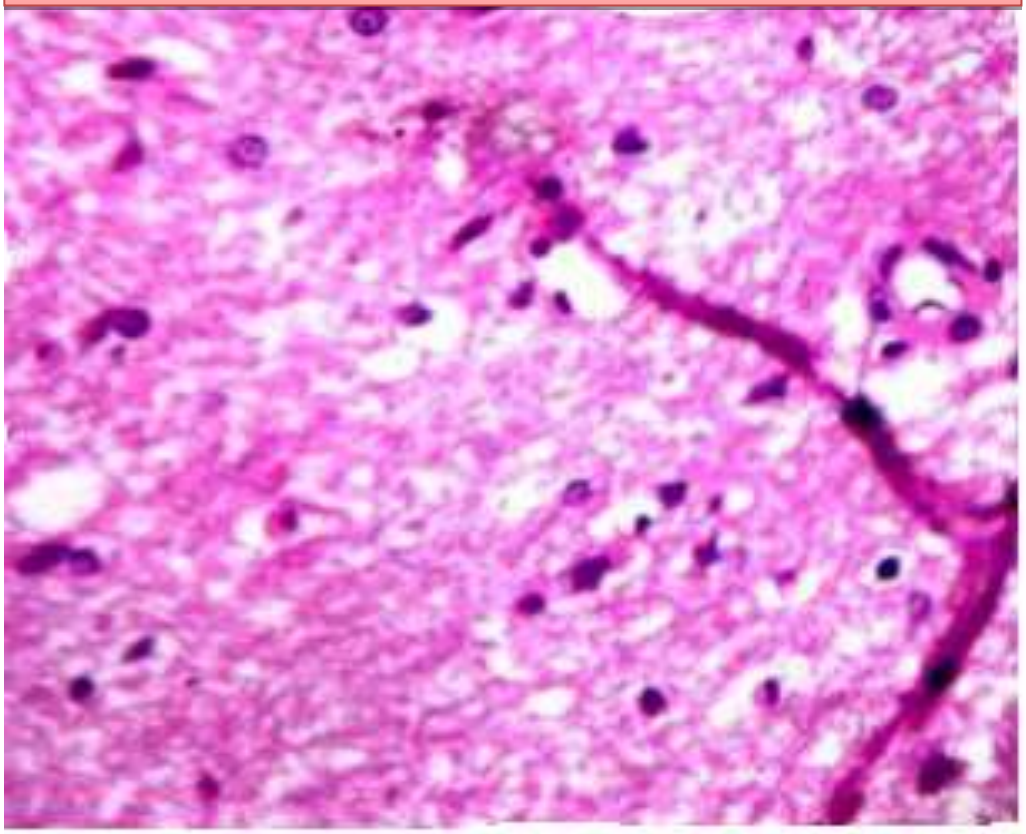

Figure5:Histopathological section of brain of rat treated with arsenic show degeneration and necrosis of neoron.X40 H&E(47).

CHAPTER THREE

CONCLUSIONS

***CONCLUSION*:**

1. Exposure to heavy metals as general and arsenic as special causes common health problems overworld due to drinking of contaminated food, air,and water.
2. Arsenic is organic, inorganic element distributed innature.
3. The arsenic is toxic heavy metal than others arsenic compounds.
4. Arsenic toxicity causes loss of abilityto interact with sulfhydryl groups of enzymes and deactivated of some enzyme such as cellular respiration, that results in inhibition of Krebs cycle and other enzymes.
5. The arsenic exposure causes increasing the oxidative stress by inhibition of antioxidants and creating Reactive Oxygen Species,also result in damage to kidney and liver tissue.
6. The main pathological lesion in theliver is vaculation of hepatocyte, necrosis,and presence of inflammatory cells. While in kidneys there is degeneration of epithelial layer of tubules and degenerative changes in brain tissu**es**

CHAPTER FOUR

RECOMMENDATION

Recommendation:

1- Decrease uses ofpesticides which contain thearsenic compound.
2- Decrease uses of drugs and cosmetic materials which have arsenic in thetreatment of special diseases.
3- Decrease the sours of contamination of water and food with arsenic to decrease poisoning cases in human and animals.
4- Make further study on another organ to show changes which occur in artery and muscles.

CHAPTER FIVE

REFERENCES

References:

1-Chen S, Chen C, Kuo C, Huang C, Lin C, Lu Z, et al. Elevated risk of hypertension induced by arsenic exposure in Taiwanese rural residents: possible effects of manganese superoxide dismutase (MnSOD) and 8-oxoguanine DNA glycosylase (OGG1) genes. Arch Toxicol. 2012;8(6):869-78. doi: 10.1007/s00204-011- 0797-8.

2-Agency for Toxic Substances and Disease Registry-ASTDR. Toxicological profiles for arsenic. American publishing press. Atlanta. The USA. 2005; pp:228.

3-Kimura A, Ishida Y, Nosaka M, Kuninaka Y, Hama M, Kawaguchi T, et al. Exaggerated arsenic nephrotoxicity in female mice through estrogen-dependent impairments in the autophagic flux. Toxicology. 2016;339:9-18. doi: 10.1016/j.tox.2015.11.005.

4-Khalil I, Abdul H, Abdul F. Dose-response relationship between arsenic exposure and the serum enzymes for liver function test in the individuals exposed to arsenic: Environ. Health J. 2011; 10 (64):102-115.

5. Droge W. Free radicals in the physiological control of cell function. Rev. 2002;82:47–95.

6. Sarkar M, Chaudhuri GR, Chattopadhyay A, Biswas NM. Effect of sodium arsenite on spermatogenesis, plasma gonadotrophins and testosterone in rats. Asian J Androl. 2003;5:27–31.

7. Ramos O, Carrizales L, Yáñez L, Mejía J, Batres L, Ortíz D, et al. Arsenic increased lipid peroxidation in rat tissues by a mechanism independent of glutathione levels. Environ Health Perspect. 1995;103:85–8

8. Li Z, Piao F, Liu S, Wang Y, Qu S. Subchronic exposure to arsenic trioxide-induced oxidative DNA damage in kidney tissue of mice. ExpToxicolPathol. 2010;62:543–547.

9. Chowdary R, Dutta A, Chaudhari SR, Sharma N, Giri AK, Chaudari K. In vitro and in vivo reduction of sodium arsenite-induced toxicity by aqueous garlic extract. Food ChemToxicol. 2008;46:740–51.

10.Michael F. Hughes Barbara D. Beck Yu Chen Ari S. Lewis David J. Thomas Arsenic Exposure and Toxicology: A Historical Perspective Toxicological Sciences, Volume 123, Issue 2, 2011;305–332.

11. Scheindlin. The duplicitousnature of inorganic arsenic, Mol. Interv. , 2005, vol. 5: 60-64.

12. Antman K. The history of arsenic trioxide in cancer therapy, Oncologist, 2001, vol. 2: 1-2.

13. Sabina C. Grund, KunibertHanusch, Hans Uwe Wolf "Arsenic and Arsenic Compounds"in Ullmann's Encyclopedia of Industrial Chemistry,VCHWiley,2008,Weinheim.doi:10.1002/14356007.a03_113

14. . Lide, DR (ed.). CRC Handbook of Chemistry and Physics. 81st Edition. CRC Press LLC, Boca Raton: FL 2000, p. 4-44.

15. Emsley, John (2001). Nature's Building Blocks: An A-Z Guide to the Elements. Oxford: Oxford University Press.

16. Domingo JL. Prevention by chelating agents of metal-induced developmental toxicity. ReprodToxicol. 1995;9:105–13.

17. Klaassen CD. Heavy metals and heavy metal antagonist. In: Gilman AG, Rall TW, Nies AS, Taylor P, editors. The Pharmacological Basis of Therapeutics. 9th ed. New York: McGraw Hill; 1996. pp. 1592–614.

18. Ratnaike RN. Acute and chronic arsenic toxicity. Postgraduate Med J. 2003;79:391–6.

19.Lindberg AL, Kumar R, Goessler W, Thirumaran R, Gurzau E, Koppova K, et al. Metabolism of low-dose inorganic arsenic in a central European population: influence of sex and genetic polymorphisms. Environ Health Perspect. 2007;115:1081–6.

20. Pal S, Chatterjee AK. Protective effect of methionine supplementation on arsenic-induced alteration of glucose homeostasis. Food and Chemical Toxicology. 2004;42:737–42.

21. Brinkel J, Khan MH, Kraemer A. A systematic review of arsenic exposure and its social and mental health effects with special reference to Bangladesh. Int J Environ Res Public Health. 2009;6:1609–19.

22. Ratnaike RN. Acute and chronic arsenic toxicity. Postgraduate Med J. 2003;79:391–6.

23. Hansen ES. International Commission for Protection against Environmental Mutagens and Carcinogens. ICPEMC Working Paper 7/1/2. Shared risk factors for cancer and atherosclerosis-a review of the epidemiological evidence. Mutat Res. 1990;239:163–79.

24. Tseng WP, Chu HM, How SW, Fong JM, Lin CS, et al. (1968) Prevalence of skin cancer in an endemic area of chronic arsenicism in Taiwan. J Natl Cancer Inst 40: 453-463.

25. Cebrian ME, Albores A, Aguilar M, Blakely E Chronic arsenic poisoning in the north of Mexico. Hum Toxicol. (1983) 2: 121-133.

26. Vahidnia A, van der Straaten RJ, Romijn F, van Pelt J, van der Voet GB, et al. Arsenic metabolites affect the expression of the neurofilament and tau genes: an in-vitro study into the mechanism of arsenic neurotoxicity. Toxicol In Vitro(2007) 2: 1104-1112.

27. Jolliffe DM, Budd AJ, Gwilt DJ Massive acute arsenic poisoning. Anaesthesia (1991) 46: 288-290.

28. Pershagen G (1983) The epidemiology of human arsenic exposure. In: BA Flower (ed.) Biological and environmental effects of arsenic (1stedn.) Elsevier, Amsterdam.

29. He W, Greenwell RJ, Brooks DM, Calderon-Garciduenas L, Beall HD, et al. (2007) Arsenic exposure in pregnant mice disrupts placental vasculogenesis and causes spontaneous abortion. ToxicolSci 99: 244-253.

30. Sanghamitra S, Hazra J, Upadhyay SN, Singh RK, Amal RC (2008) Arsenic-induced toxicity on the testicular tissue of mice. Indian J PhysiolPharmacol 52: 84-90.

31. Santra A, Maiti A, Das S, Lahiri S, Charkaborty SK, et al. (2000) Hepatic damage caused by chronic arsenic toxicity in experimental animals. ToxicolClinToxicol 38: 395-405.

32. Gora RH, Baxla SL, Kerketta P, Patnaik S, Roy BK (2014) Hepatoprotective activity of Tephrosiapurpurea against arsenic-induced toxicity in rats. Indian J Pharmacol 46: 197-200.

33. Zaldívar R (1980) A morbid condition involving cardio-vascular, bronchothoracic , digestive and neural lesions in children and young adults after

34. Gora RH, Baxla SL, Kerketta P, Patnaik S, Roy BK (2014) Hepatoprotective activity of Tephrosiapurpurea against arsenic-induced toxicity in rats. Indian J Pharmacol 46: 197-200.

35. Bruguera M, Aranguibel F, Ros E .Incidence and clinical significance of sinusoidal dilatation in liver biopsies. Gastroenterol; (1978) 75: 474-8.

36.Singh AP, Goel RK, Kaur T.MechanismsAbout Arsenic Toxicity. ToxicolInt(2011) 18: 87- 93.

37. D.N. GuhaMazumder, in Handbook of Arsenic Toxicology, 2015Health Effects Chronic Arsenic Toxicity

38. NamdarYousofvand, Mohammad Fahim: Effects of Chronic Exposure to Sodium Arsenate on Kidney of Rats. Iranian Journal of Toxicology Volume 9, No 30, 2015.

39. Li, Z., F. Piao, S. Liu, Y. Wang and S. Qu, Subchronic exposure to arsenic trioxide-induced oxidative DNA damage in kidney tissue of mice. Exp. Toxicol. Pathol., 62: 543–557:2010.

40.Vijay Kumar, Chandra Prakash,ManishaSoni,Mitochondrial oxidative stress and dysfunction in arsenic neurotoxicity: A review. Journals of applied toxicology(2015).

41. Koehler Y, Luther EM, Meyer S, Schwerdtle T, Dringen R, et al. (2014) Uptake and toxicity of arsenite and arsenate in cultured brain astrocytes. J Trace Elem Med Biol 28: 328-337.

42. Rai NK, Ashok A, Rai A, Tripathi S, Nagar GK, et al. (2013) Exposure to As, Cd and Pb-mixture impairs myelin and axon development in rat brain, optic nerve and retina. ToxicolApplPharmacol 273: 242-258.

43. Kim M, Seo S, Sung K, Kim K (2014) Arsenic exposure in drinking water alters the dopamine system in the brains of C57BL/6 mice. Biol Trace Elem Res 162: 175-80.

44. Chandravanshi LP, Shukla RK, Sultana S, Pant AB, Khanna VK (2014) Early life arsenic exposure and brain dopaminergic alterations in rats. Int J Dev Neurosci 38: 91-104.

45. Ghosh A, Mandal AK, Sarkar S, Das N (2011) Hepatoprotective and neuroprotective activity of liposomal quercetin in combating chronic arsenic-induced oxidative damage in liver and brain of rats. Drug Deliv 18: 451-459.

46. Oluwole Israel Oyewole1, TolulopeOlufunmilayoAkinbamijo, Oluwabukola Joy Omoboriowo, Naomi Ubongabasi Ukana1 1Department of Biochemistry, Osun State Hepatorenal Protective Activities of Plantain Root (Musa paradisiaca) on Arsenic-induced Oxidative Damage in Rat Advances in Biochemistry 2015; 3(2): 30-34

47. Mukesh Kumar Mundey, Manju Roy, Sushovan Roy, Manoj Kumar Awasthi, Richa Sharma. Antioxidant Potential of Ocimum sanctum in Arsenic-Induced Nervous Tissue Damage. Braz J Vet Pathol, 2013, 6(3), 95 – 101.